JVI Journals.ASM.org

GENOME ANNOUNCEMENT

Complete Genome Sequence of a New Circular DNA Virus from Grapevine

Björn Krenz,[a] Jeremy R. Thompson,[a] Marc Fuchs,[b] and Keith L. Perry[a]

Department of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, New York, USA,[a] and Department of Plant Pathology and Plant-Microbe Biology, Cornell University, New York State Agricultural Experiment Station, Geneva, New York, USA[b]

A novel circular DNA virus sequence is reported from grapevine. The corresponding genomic organization, coding potential, and conserved origin of replication are similar to those of members of the family *Geminiviridae*, but the genome of 3,206 nucleotides is 4% larger than the largest reported geminiviral genome and shares only 50% overall sequence identity.

Geminiviruses are plant viruses with geminate icosahedral particles and a circular single-stranded DNA genome, among other features. Their recent emergence is notable based on a rapidly expanding geographic distribution and host range, as well as a recombination propensity that can cause new diseases and new epidemics (4, 6). We provide here the first description of a geminilike virus sequence from grapevine (*Vitis vinifera*), for which the corresponding virus is provisionally named grapevine cabernet franc-associated virus (GCFaV).

Circular DNA was amplified from grapevine total nucleic acid extracts by rolling circle amplification (RCA) and resolved by restriction fragment length polymorphism (RFLP) (2); the products were detected from four different *Vitis vinifera* 'Cabernet Franc' vines, all originating from the same declining vineyard in New York. No comparable DNA was detected in 18 other grapevine samples collected from different sites, including two 'Cabernet Franc' accessions from independent sources. Cloning and Sanger sequencing (4× coverage, both strands; Vector NTI software assembly) revealed a single DNA circle of 3,206 nucleotides (nt). This genome size is larger than the largest previously reported geminivirus genome of 3,080 nucleotides (3).

Consistent with other monopartite members of the family *Geminiviridae*, the orientation of the predicted GCFaV open reading frames (ORFs) is bidirectional, with three ORFs in the viral-sense orientation (V) and three in the complementary orientation (C). Importantly, the nonanucleotide signature for the geminivirus origin of replication, TAATATT|AC, was present in an intergenic region, as observed in all members of the family *Geminiviridae* (5). BLASTN analysis (1) showed the closest related sequence to be that of a dicot-infecting mastrevirus, chickpea chlorotic dwarf Syria virus, the genome of which is 634 nt smaller and shares only 50% identity. The GCFaV ORF V1 (coat protein [CP]) showed a maximum amino acid sequence identity of 26% with *Mesta yellow vein mosaic virus* (genus *Begomovirus*). Remarkably, the V2 and V3 ORFs had no apparent sequence similarity with other geminiviral sequences at the nucleotide and amino acid levels. The ORFs C1 and C2 showed a subgenomic organization strikingly similar to those of mastreviruses (including a spliced transcript) (7) and maximum identities of 33% and 52%, respectively, with 74% and 79% coverage (BLASTX analysis) of the respective ORFs of *Bean yellow dwarf virus*. In phylogenetic analyses and maximum likelihood and neighbor-joining trees for the CP gene, replicase gene, and full-length sequences, GCFaV formed a distinct branch (bootstrap > 70%) apart from all members of four genera within the family *Geminiviridae*.

A DNA virus belonging to the genus *Badnavirus*, family *Caulimoviridae*, was recently detected in grapevine by deep sequencing (8), but this is the first report of a geminivirus sequence in grapevine. Further studies are needed to determine the prevalence of GCFaV and its impact.

Nucleotide sequence accession number. The GenBank accession number for the sequence of GCFaV is JQ901105.

ACKNOWLEDGMENTS

This work was supported in part by funding from the U.S. Department of Agriculture Animal Plant Health Inspection Service as part of the National Clean Plant Network and by Agriculture and Food Research Initiative Competitive Grant no. 2009-55605-05184 from the USDA National Institute of Food and Agriculture.

REFERENCES

1. **Altschul SF, et al.** 1997. Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. Nucleic Acids Res. **25**:3389–3402.
2. **Haible D, Kober S, Jeske H.** 2006. Rolling circle amplification revolutionizes diagnosis and genomics of geminiviruses. J. Virol. Methods **135**:9–16.
3. **Klute KA, Nadler SA, Stenger DC.** 1996. Horseradish curly top virus is a distinct subgroup II geminivirus species with rep and C4 genes derived from a subgroup III ancestor. J. Gen. Virol. **77**:1369–1378.
4. **Moffat AS.** 1999. Plant pathology—geminiviruses emerge as serious crop threat. Science **286**:1835.
5. **Stanley J, et al.** 2005. Geminiviridae, p 301–326. *In* Ball LA (ed), Virus taxonomy. Elsevier/Academic Press, London, United Kingdom.
6. **Varma A, Malathi VG.** 2003. Emerging geminivirus problems: a serious threat to crop production. Ann. Appl. Biol. **142**:145–164.
7. **Wright EA, Heckel T, Groenendijk J, Davies JW, Boulton MI.** 1997. Splicing features in maize streak virus virion- and complementary-sense gene expression. Plant J. **12**:1285–1297.
8. **Zhang Y, Singh K, Kaur R, Qiu W.** 2011. Association of a novel DNA virus with the grapevine vein-clearing and vine decline syndrome. Phytopathology **101**:1081–1090.

Received 16 April 2012 Accepted 24 April 2012

Address correspondence to Keith L. Perry, KLP3@cornell.edu.

doi:10.1128/JVI.00943-12